

investigations in these laboratories are confirmatory of the results of the investigations in the laboratories heretofore mentioned. A special investigation is being conducted, however, in the Hoagland Laboratory by Dr. E. H. Wilson, upon the life-history of the bacillus, and in a telegram received February 14th, in response to an inquiry, he informs me that the bacillus in a dark closet, on filter-paper and on wool-blanket, is still living at the end of the eighteenth day. The experiments conducted by Drs. J. J. Kinyoun and H. D. Geddings in the Laboratory of the Marine-Hospital Service have related especially to the action of disinfectants, the results of which are set forth in the preceding table.

The prescribed limits of this article prevent any considerations concerning the contagious and infectious character of the disease and the conditions which cause its spread. Neither is it possible herein to describe the measures which have been taken to prevent the spread of the disease in foreign countries and to prevent its introduction into the United States. These subjects would require a special treatise.

THE SERUM-TEST FOR THE DIAGNOSIS OF TYPHOID FEVER,

WITH A DESCRIPTION OF THE METHODS FOLLOWED AND THE RESULTS
OBTAINED IN ITS USE IN THE LABORATORIES OF THE HEALTH
DEPARTMENT OF NEW YORK CITY.

BY HERMANN M. BIGGS, M.D.,
PATHOLOGIST AND DIRECTOR OF THE BACTERIOLOGICAL LABORATORIES,

AND

WILLIAM H. PARK, M.D.,
ASSISTANT DIRECTOR, HOSPITAL BACTERIOLOGICAL LABORATORY.

As has been pointed out by Widal, the observations of Pfeiffer, Grüber, and Widal, showing that the blood-serum of persons ill with typhoid fever exercises a peculiar influence upon recent broth-cultures of the typhoid bacillus, introduce an entirely new phase in the practical application of bacteriological work in the study of the infectious diseases.

Hitherto bacteriological investigations, in their clinical application, have been confined solely to the demonstration of pathogenic micro-organisms, either by culture or by direct microscopical examination in the tissues, fluids, or secretions of the body. These new observations have disclosed a method which permits of the diagnosis of typhoid fever by the peculiar reaction occurring in broth-cultures of the typhoid bacillus when the blood-serum of a typhoid patient is added.

The development of our present knowledge of the subject may be briefly traced as follows :

As long ago as 1892 Grüber described an agglomeration of the bacilli as occurring in guinea-pigs which had died after cholera-infection; but to Pfeiffer is undoubtedly due the credit of having observed and fully described the peculiar influence of the respective serums of immunized animals on the cholera-vibrium and the typhoid bacillus when added to broth-cultures of these organisms and the mixtures then introduced into the peritoneal cavity of other animals.

In various communications published in 1894 and 1895 Pfeiffer showed that when cultures containing dead or living cholera-spirilla or typhoid bacilli are injected subcutaneously into animals or man specific protective substances are formed in the blood of the individuals thus treated. These substances grant a more or less complete immunity against the invasion of the living germs of the respective diseases. He also described the occurrence of a peculiar phenomenon when a portion of a fresh culture of the cholera-vibrium on agar is added to a small quantity of the serum of an animal immunized against cholera, and the mixture injected into the peritoneal cavity of a non-immunized guinea-pig. After this procedure, if from time to time minute drops of the liquid be withdrawn in capillary tubes and examined microscopically, it is found that the vibrios, which were formerly, and in control-animals which remain, actively motile, become in a very short time, under the influence of the serum, entirely motionless and later dead. They are first immobilized, then they become somewhat swollen and agglomerated into balls or clumps, which gradually become paler and paler, until finally they are dissolved in the peritoneal fluid. This process takes place regularly, in about twenty minutes, provided a sufficient degree of immunity be present in the animals from which the serum was obtained.

The animals injected with the mixture containing cholera-cultures and the serum of immunized animals remain unaffected, while control-animals treated with a fluid containing only the serum of non-immunized animals mixed with cultures of the cholera-vibrium promptly die.

Pfeiffer claimed that the reaction of the serum employed in this manner is so distinctly specific that it may serve for the differential diagnosis of the cholera-vibrium from other vibrios. If a mixture of the cultures of two vibrios is used in this experiment, only that species is destroyed which has been employed to immunize the animal whose serum is introduced with the cultures. These latter observations of Pfeiffer were made before 1894, and were demonstrated to one of the writers early in 1894 in the Institute for Infectious Diseases in Berlin.

In March, 1896, Pfeiffer and Kolle published an article in the *Deutsche med. Wochenschrift*, entitled "The Differential Diagnosis of Typhoid Fever by Means of the Serum of Animals Immunized against Typhoid Infection." In this communication the authors say that by the aid of the serum of convalescents from typhoid fever, or of animals immunized

against typhoid infection, typhoid bacilli can be differentiated from the bacillus coli communis and from other bacteria resembling the typhoid bacillus. This is accomplished by mixing the serum with cultures of the typhoid bacillus and introducing the mixture into the peritoneal cavity of guinea-pigs. A specific reaction appears, which can be followed microscopically. Agglomeration and agglutination of the bacilli occur, followed by deformity and final solution. The animals remain unaffected. The bacteria belonging to the coli group, and all other similar bacteria, are not influenced in this test when the serum of animals immunized to the typhoid bacillus is employed. "This change takes place only with the typhoid bacilli, and is a specific reaction due to the bactericidal reaction of the typhoid serum."

It was further found, if the serum from a goat, which had been immunized (so that its immunity was about ten times as great as that of the average convalescent from typhoid fever), was diluted with forty parts of bouillon, and a similar dilution made of the serum of non-immunized animals, and both solutions were then inoculated with a culture of the typhoid bacillus and placed in an incubator at 37° C., that after the expiration of one hour macroscopical differences in the culture could be observed, which increased in distinctness for four hours and then gradually disappeared. The reaction occurring may be described as follows: in the tube in which the typhoid culture is mixed with typhoid serum the bacilli are agglomerated in fine whitish flakes, which settle to the bottom of the tube, while the supernatant fluid is clear or only slightly cloudy. On the other hand, the tubes containing mixtures of bouillon with the cholera-serum, or the serum from non-immunized animals inoculated with the typhoid bacillus, became and remained uniformly and intensely cloudy. These two serum-mixtures, examined microscopically in a hanging drop, also show distinct differences. The typhoid serum mixture inoculated with the typhoid bacilli exhibits the organisms entirely motionless, lying clumped together in heaps; in the other mixture the bacilli are actively motile.

Pfeiffer and Kolle say further: "No such differences were shown when bacteria of the coli group were examined after mixture with the typhoid serum." They conclude: "It would seem, therefore, that this method, which is simple of execution in any laboratory, may be of great practical value as an aid to differential diagnosis."

Grüber, in an article in the *Wiener med. Wochenschrift* for March, 1896, describes some investigations conducted in conjunction with Dr. Herbert E. Durham, of London. He criticises Pfeiffer's conclusions and attempts to show by a series of experiments that the reaction described by Pfeiffer is by no means specific, and that when the reaction is positive the diagnosis still remains in doubt, for the reaction is *quan-*

titative only, and not qualitative, so far as the cholera-vibron is concerned. Grüber expresses the opinion, however, that it is of greater service with regard to the typhoid bacillus, and concludes that these investigations will render great assistance in the clinical diagnosis of cholera and typhoid fever. He describes a method for applying the test in regard to both of these organisms, which closely resembles that of Pfeiffer's.

Durham published a paper in the *British Medical Journal* of March 19, 1896, on "The Special Reaction of the Serum of Highly Immunized Animals, and the Use of this Serum for Diagnostic and Other Purposes." He describes in considerable detail the use of these serums, and draws a series of conclusions as to the value of the reactions.

The first practical application of this method for the diagnosis of typhoid fever on a more extensive scale was made by Widal and reported with great fulness and detail in a communication published in June, 1896, in *La Semaine Méd.*

Widal describes the phenomenon as follows: "Several bouillon-tubes are separately inoculated with the typhoid bacillus and the bacillus coli communis. A few drops of the serum of an animal thoroughly immunized against typhoid infection are added to each of these, and they are then placed in an incubator at 37° C. In four or five hours the tubes containing the bacillus coli communis begin to become cloudy, while the tubes inoculated with the typhoid bacillus remain almost entirely clear. At the end of twenty-four hours the tubes with the bacillus coli have become intensely cloudy throughout, while those containing the typhoid bacillus remain clear or are but slightly cloudy, the microbes in these tubes having been precipitated as whitish flakes to the bottom.

"A drop of the culture of the bacillus coli examined microscopically shows isolated bacteria, for the most part actively motile. A drop of the culture of the typhoid bacillus, on the other hand, when examined microscopically, presents only scattered agglomerations of bacteria which are immobile, deformed, thickened, and agglutinated together. No isolated or motile bacteria are to be seen. The formation of a precipitate visible to the naked eye, and, microscopically, the immobilization, agglutination, and deformity of the microbes are the characteristics of the phenomenon.

"The serum of an animal thoroughly immunized to the typhoid bacillus is more active than that of a patient suffering with or convalescing from typhoid fever. The greater the immunization to which the animal has been subjected the more active is the serum.

"By the use of this test," Widal continues, "I have examined the serum obtained from some six typhoid fever patients on the seventh, twelfth, fifteenth, sixteenth, nineteenth, and twenty-first days of the disease, to determine whether it could be employed for the clinical diagnosis of

typhoid fever. Each time I readily obtained undoubted evidences that the serum exerted an immobilizing and agglutinating action on the bacilli in cultures of the typhoid organism."

The method employed in this investigation was as follows: a small quantity of blood having been withdrawn, under aseptic precautions, with a sterilized syringe from a vein at the elbow, the serum, after coagulation of the blood, is decanted and a few drops introduced into a tube containing a broth-culture of the typhoid bacillus, the proportion being one part of serum to ten or fifteen parts of broth. The tube is then placed in an incubator at a temperature of 37° C. After twenty-four hours, when the typhoid serum has been employed, the characteristic reaction as described is apparent to the naked eye, and is seen microscopically in the hanging drop.

The peculiar appearance of the culture is still evident after two, three, or four days, but at the end of the first twenty-four hours the precipitate seems to be most abundant.

"This phenomenon may be observed in a simpler manner," Widal says, "by withdrawing several drops of blood from the end of the finger, under aseptic precautions, and after the blood has clotted the serum is separated and added drop by drop to a broth-culture, in the proportion of one to ten. If the serum has been obtained from a case of typhoid fever, a characteristic reaction will be immediately observed.

"When the blood-serum of patients in good health, or of those suffering from other diseases than typhoid fever, is subjected to this test, few or none of the appearances observed in the above reaction are seen. The reaction may be found for considerable periods after recovery from typhoid fever."

In subsequent communications Widal reports further observations. In one of these communications he refers to the resistance of the agglutinating property of the serum, and notes that the dried serum and the dried blood as well, to a less extent, retain the specific action.

Achard has shown that exposure to diffused sunlight, extending over a period of weeks, produces no change in the agglutinating property of the blood or serum. He also found that various fluids and secretions of typhoid patients possessed the same property in a greater or less degree. The serous fluid of blisters, the serum from pus, tears, and urine of typhoid patients; the milk of a wetnurse sick with typhoid fever, and other fluids possess this property. It was found that in the inflammatory exudations of serous membranes occurring during typhoid fever this property was present when the exudate did not occur too rapidly.

Achard also found the same to be true with regard to the secretion of the lachrymal glands. If the secretion was taken with a pipette from the inner canthus of the eye, it gave a marked reaction; but if the secre-

tion was markedly increased by inhaling ammonia-vapor, the rapidly flowing tears did not give the reaction.

Achard and Bensaude found that the milk of a wetnurse suffering from typhoid fever, which gave a reaction, lost this property when the milk was not pure and freshly drawn. They also found that prolonged exposure to a temperature of 60° C. did not affect the reaction; but that it was considerably diminished at 100° C., and was lost after boiling the milk for fifteen minutes at 120° C. It also disappeared after filtration through a porcelain tube, and the same was found to be the case with urine. The blood-serum of a sucking-infant, fed with milk which showed the reaction, did not possess it.

Widal and Achard attempted to ascertain what substances in the blood possess the agglutinating power. They found that blood-serum deprived of globulin had lost the agglutinating property, while the globulin retained it. They found also that when globulin and fibrin have been separated from the blood and dissolved, that each one of these substances possesses intense agglutinating power; while the plasma deprived of these, and containing only albumin, has lost its specific action. The same has been found to be true of the serous fluid of blisters and the serous fluid of the pericardium, peritoneum, etc. It would appear, therefore, that the agglutinating property belongs to the fibrin and the globulin of the blood. Similar results were noted in relation to the albumin and globulin of milk. Casein isolated and dissolved preserves powerful agglutinating properties.

The observations of Widal and others would seem to show that this peculiar phenomenon is not a reaction of immunization, but is "an infection-reaction," becoming less pronounced and sometimes disappearing during convalescence; that is, diminished at the time when immunity is most marked. In some cases it has not been found on one day, and has appeared on the next; or it is present in marked form on one day, and has disappeared on the next. In the majority of cases it diminishes in intensity soon after convalescence is established and frequently soon disappears; while in other persons it remains fairly distinct, even for some years after recovery. The serum does not actually destroy all the bacteria, as is shown by the fact that cultures still retain their power of growth after transplantation, even when mixed with active typhoid serum in equal parts and allowed to remain for long periods exposed to its action.

All observers have agreed in confirming the observations of Pfeiffer, Grüber, and Widal as to the occurrence of this peculiar phenomenon in most cases of typhoid fever at some period in the course of the disease; but in some instances it appeared on the fourth or fifth day, in others not until the end of the third week or later, and rarely not at all.

TABLE

giving the results of the method of sero-diagnosis in cases of clinically typical or suspected typhoid fever, in convalescents or those who have had typhoid, in diseases other than typhoid, and in healthy subjects, showing the number of cases tested, the period when the test was made, and the reaction.

Reporter.	Number of cases of typical or suspected typhoid tested.	Day of disease when tested.	Reaction.			Number of convalescents tested.			Number of cases other than typhoid tested.		Number of healthy subjects tested.		Test used.		
			Positive.		Negative.	Reaction.		Period when tested after convalescence.	Reaction.		Post- negative.	Nega- tive.			
			Decis- ive.	Partial or feeble.	Decisive or firm.	Typheid or no reaction.	Post- negative.	Nega- tive.	Post- negative.	Nega- tive.					
Widal,	80	4th to 21st day.	45		35			6	10	0	200	0	39	Fluid serum.	
Achard,	9	7th to 12th day.	8		3	8								" "	
Dieulafoy,	2	7th to 12th day.	2											" "	
Chantemesse,	11	9th day.	11								0	No. not stated.		" "	
Rendu,	1	11th day.	1											" "	
Haushalter,	39	7th to 12th day.	27		12									" "	
Veiel,	2		1		1									" "	
Courmont,	13		11		2									" "	
Gatrin,	57	4th to 41st day.	36		21					0	20			" "	
Theolen and Mills,	12		12											" "	
Grünbaum,	8	10th to 33d day.	8								15 ¹	4	4 ¹	3	" "
Durham,	4	8th to 21st day.	2			2								" "	
Delépine,	37	8th to 23d day.	24	1	4						0	10		" and dried blood.	
Greene,	11	16th to 38th day.	11								0	14		Fluid serum.	
Johnston,	143	48 hrs. to 3d wk.	118	5	14						0	85		Dried blood.	
Total,	422	2d dy. to 4th wk.	312	0	92	12					15	203	4	42	

¹ The positive reactions obtained by Grünbaum in cases, other than typhoid and in healthy subjects, occurred only when the serum was used in greater concentration than 1 to 10. In mixtures of 1 to 10 the reaction occurred only in typhoid fever.

The table on page 280, prepared by Dr. Arthur R. Guerard, Assistant Bacteriologist to the Health Department, shows, as well as is possible in tabular form, the results reported by various observers up to January 1, 1897, in the use of the serum-test for the diagnosis of typhoid fever.

In a total of 422 cases of typical or suspected typhoid fever, reported by fifteen different observers, 404 were diagnosticated decisively to be typhoid fever or not, 312 positively and 92 negatively; or in about 95 per cent. of cases the diagnosis was confirmed by the serum-test on the first examination; 6 of the positive reactions were partial or feeble at first, but on re-examination were decisive. 12 of the negative reactions were doubtful or remained indecisive.

In a total of 63 cases who had had typhoid fever, 32 gave the reaction and 30 did not. In 308 cases of diseases other than typhoid there was no reaction with the blood or serum in a dilution of 1 part of serum to 9 of the culture, but when the dilution of serum was less than 1 to 9 a reaction frequently occurred. Of 46 healthy subjects examined, 4 gave the reaction, or about 10 per cent.; but many persons, the number of which is not stated in the reports, were examined and gave no reaction with the typhoid cultures; therefore these data are incomplete.

THE USE OF THE SERUM-TEST IN THE LABORATORIES OF THE NEW YORK HEALTH DEPARTMENT.

Wyatt Johnston, of Montreal, introduced the use of this method in municipal laboratories for the diagnosis of typhoid fever, suggesting that dried blood should be employed in place of blood-serum. Widal had shown that drying did not destroy the agglutinating properties of typhoid blood. In September and October, 1896, some investigations on this subject were begun in the laboratories of the Health Department of New York City, and in the latter part of October the Health Board issued an announcement to the physicians of this city, from which the following extracts are made:

"Observations on the blood-test for the diagnosis of typhoid fever, which have been in progress in the laboratories of the New York Health Department for some time past, have thus far confirmed the conclusions of previous investigators, and it seems probable that we have in this method not only a rapid, but also a reliable diagnostic test. Comparatively few cases, however, have as yet been examined very early in the course of the disease, and it is not positively known how early the reaction ordinarily appears.

"The Health Department therefore requests that physicians having cases which present symptoms suggestive of typhoid fever send specimens of the blood daily, or every other day, until the nature of the disease shall clinically be clearly defined.

"One source of error which must be guarded against arises from the persistence of the reaction in some cases for a number of years after an attack of typhoid fever, so that a reaction may appear in health, or in affections other than typhoid fever, if the individual have previously suffered from the latter disease.

"It must be distinctly understood that the observations thus far made are not sufficiently numerous and complete to permit a positive conclusion as to the exact value or the limitations of this test, and it is the desire of the Health Department to supply as soon as possible the data necessary to such conclusion.

"When a negative result is reported in a suspicious case the physician in attendance is particularly requested to send further specimens, and in every case to notify the Director of the Laboratory as to whether the laboratory diagnosis is finally in harmony with the clinical diagnosis or is at variance with it. In the cases in which the reaction is decided it may apparently be stated positively that the patient either has, or has had within a few years, typhoid fever. In those cases in which the reaction is present, but not well marked, only a probable diagnosis can be made. The complete absence of the reaction in the blood of a patient sick more than one week (if further examinations confirm the results thus far obtained) will justify the exclusion of the diagnosis of typhoid fever.

"DIRECTIONS FOR PREPARING THE SPECIMENS OF BLOOD. The skin covering the tip of the finger is thoroughly cleansed and is then pricked with a clean needle deeply enough to cause several drops of blood to exude. Two drops are then placed on the accompanying glass slide, one near either end. After drying, the slide is placed in the holder and returned to the station from which it was obtained or mailed to the laboratory. Full data of the case must be forwarded with each specimen. A report of the result of the examination will be mailed to the attending physician on the following day."

Arrangements were at once made so that physicians could obtain glass slides for collecting blood, with slide-holders, circulars of information, and the accompanying blanks to be filled out, from the various health department depots (nearly one hundred in number) scattered through the city. After preparation of the specimens they are returned to one of the depots and are collected by employ  s of the health department each afternoon, with the specimens of sputum for the diagnosis of tuberculosis and culture-tubes for the diagnosis of diphtheria. Copies of the blanks follow:

88 L-1896.

BLANK. FIRST SPECIMEN.

Typhoid Fever.

See other side for directions for preparing specimens and for temperature-record.

Date Time
 Name of Patient Age
 Address Occ
 Attending Physician
 Address
 Clinical Diagnosis
 Duration of Illness
 Has Patient previously had Typhoid Fever?
 If so, how long ago?
 If Typhoid Fever, where contracted?
 Has Patient been outside of New York City during the month previous to present illness?
 If so, where?

This blank should be filled out in full by attending physician.

BLANK. FIRST SPECIMEN (REVERSE SIDE).

Directions for Preparing Specimens of Blood.

The skin covering the tip of the finger is thoroughly cleansed and is then pricked with a clean needle deeply enough to cause several drops of blood to exude. Two drops are then placed on the accompanying glass slide, one near either end. After drying, the slide is placed in the holder and returned to a culture-station, or mailed to the laboratory. The accompanying blank must be fully filled out and forwarded with the specimen. A report of the result of the examination will be mailed to the attending physician on the following day.

Communications should be addressed to

HERMANN M. BIGGS, M.D.,
 Pathologist and Director of the Bacteriological Laboratories,
 Criminal Court Building, New York City.

TEMPERATURE RECORD.

	1 st DAY	2 nd DAY	3 rd DAY	4 th DAY	5 th DAY	6 th DAY	7 th DAY	8 th DAY	9 th DAY	10 th DAY
	A.M. P.M.	A.M. P.M.	A.M. P.M.	A.M. P.M.	A.M. P.M.	A.M. P.M.	A.M. P.M.	A.M. P.M.	A.M. P.M.	A.M. P.M.
107°										
106°										
105°										
104°										
103°										
102°										
101°										
100°										
99°										

89 L-1896.

BLANK. LATER SPECIMEN.

Typhoid Fever. Later Specimens.

No. of Specimen : 2d, 3d, 4th, 5th, 6th, 7th, 8th.

Date Time

Name of Patient Laboratory No.

Address

Attending Physician

Address

Duration of Illness

Clinical Data noted since Previous Specimen was taken :

.....

.....

.....

.....

This blank should be filled out in full by attending physician.

See other side for directions for preparing specimens and for temperature-record.

BLANK. LATER SPECIMEN (REVERSE SIDE).

Directions for Preparing Specimens of Blood.

The skin covering the tip of the finger is thoroughly cleansed, and is then pricked with a clean needle deeply enough to cause several drops of blood to exude. Two drops are then placed on the accompanying glass slide, one near either end. After drying, the slide is placed in the holder and returned to a culture-station, or mailed to the laboratory. The accompanying blank must be fully filled out and forwarded with the specimen. A report of the result of the examination will be mailed to the attending physician on the following day.

Communications should be addressed to

HERMANN M. BIGGS, M.D..

*Pathologist and Director of the Bacteriological Laboratories,
Criminal Court Building, New York City.*

TEMPERATURE RECORD SINCE PREVIOUS SPECIMEN WAS TAKEN.

DAY DIS- EASE																		
	AM.	PM.	AM.	PM.	AM.	PM.	AM.	PM.	AM.	PM.	AM.	PM.	AM.	PM.	AM.	PM.	AM.	PM.
107°																		
106°																		
105°																		
104°																		
103°																		
102°																		
101°																		
100°																		
99°																		

The facilities thus offered to the physicians of New York City for determining the value of the serum-test for the diagnosis of typhoid fever have been quite largely utilized, and a full description of the results obtained is given below.

In a majority of the tests made in the Health Department laboratories specimens of dried blood have been used; in a small number only the serum from coagulated blood or blisters.

Ordinarily the reaction has been watched under the microscope in the hanging drop. The influence of the serum upon larger quantities of partially or completely developed broth-cultures has only been observed

in a moderate number of cases for the purpose of comparing the results with the routine method.

TECHNIQUE OF OBTAINING SERUM AND DRIED BLOOD. Blood may be easily obtained by pricking the tip of the finger or the ear. Two or three large drops should be collected on a glass slide and allowed to dry. Paper is not as good a receiver for the blood as glass, for the blood soaks more or less into it, and later, when it is dissolved, some of the paper-fibre is apt to be rubbed off with it.

In preparing the specimen for examination the dried blood is brought into solution by mixing it with about five times the quantity of water. Then a drop of this decidedly reddish mixture is placed on a cover-glass and to it is added a drop of a fifteen- to twenty-hour bouillon-culture of the typhoid bacillus. The two drops, after being mixed, should have a faint reddish tinge. The cover-glass, with the mixture on the surface, is inverted over a hollow slide (the edges about the concavity having been smeared with oil or fluid vaseline so as to make a closed chamber, and the hanging drop then examined under the microscope (preferably by gaslight), a high-power dry lens (about $\frac{1}{4}$ inch) being used.

If the reaction takes place rapidly, the first glance through the microscope reveals the completed reaction, all the bacilli being in loose clumps and nearly or altogether motionless. Between the clumps are clear spaces containing few or no isolated bacilli.

If the reaction is a little less complete, a few bacilli may be found moving slowly between the clumps, in an aimless way, while others attached to the clumps by one end are apparently trying to pull away, much as a fly caught on fly-paper struggles for freedom.

If the agglutinating substances are still less abundant, the reaction may be watched through the whole course of its development. Immediately after mixing the blood and culture together it will be noticed that many of the bacilli move more slowly than before the addition of the serum. Some of these soon cease all progressive movement, and it will be seen that they are gathering together in small groups of two or more, the individual bacilli being still somewhat separated from each other. Gradually they close up the spaces between them, and clumps are formed. According to the completeness of the reaction, either all the bacilli may finally become clumped and immobilized or only a small portion of them, the rest remaining freely motile, and even those clumped may appear to be struggling for freedom. With blood containing a large amount of the agglutinating substances all gradations in the intensity of the reaction may be observed, from those shown in a marked and immediate reaction to those appearing in a late and indefinite one, by simply varying the proportion of blood added to the culture-fluid.

PSEUDO-REACTIONS WITH DRIED BLOOD. If too concentrated a solution of dried blood from a healthy person is employed, there will be an

immobilization of the bacilli, but no true clumping. This is sometimes mistaken for a reaction. Again, dissolved blood always shows a varying amount of detritus, partly in the form of fibrinous clumps, and prolonged microscopical examination of the mixture of dissolved blood with a culture-fluid shows that the bacilli often become more or less entangled in these clumps, and in the course of one-half to one hour very few isolated motile bacteria are seen. The fibrinous clumps, especially if examined with a poor light, may be very easily mistaken for clumps of bacilli. This pseudo-reaction is regarded by many inexperienced observers as a true typhoid reaction, but it occurs as readily with non-typhoid as with typhoid blood.

THE USE OF SERUM OBTAINED FROM BLISTERS OR THE BLOOD. Fluid serum can be easily obtained in two ways: 1st. The serum may be obtained directly from the blood thus: the tip of the finger or ear is pricked with a lancet-shaped needle, and the blood as it issues is allowed to fill by gravity a capillary tube having a central bulb. The ends of the tube are then sealed by heat or wax, and as the blood clots a few drops of serum separate. This method of obtaining blood-serum has the advantage of rapidity; but also this disadvantage, namely, that the serum thus separated is apt to contain more or less blood-cells, which somewhat obscure the field when the liquid serum is immediately mixed with the culture, and which cause clumps of detritus (resembling, though less marked, those found with the dried blood), if the serum is allowed to dry. 2d. The serum may be obtained from a blister. In the examinations thus far made this has given far more satisfactory results. The method is as follows: a piece of cantharides-plaster the size of a five-cent piece is applied to the skin at some spot on the chest or abdomen. A blister forms in from six to eighteen hours. This should be protected from injury by a vaccine shield or bunion plaster. The serum from the blister is collected in a capillary tube, the ends of which are then sealed. Several drops of the serum can be easily obtained from a blister so small that it is practically painless and harmless. The serum obtained is clear and admirably suited for the test.

THE ADVANTAGES AND DISADVANTAGES OF SERUM AND DRIED BLOOD FOR THE SERUM-TEST. The dried blood is easily and quickly obtained and does not deteriorate or become contaminated by bacterial growth. It is readily transported, and seems to be of nearly equal strength with the serum in its agglutinating properties. It must, however, in use, be diluted with at least five times its bulk of water, otherwise it is too viscid to be properly employed. The amount of dilution can only be determined roughly by the color of the resulting mixture, for it is impossible to estimate accurately the amount of dried blood from the size of the drop.

Serum, on the other hand, can be used in any dilution desired, varying

from a mixture which contains equal parts of serum and broth-culture to that containing 1 part of serum to 100 parts of culture. It can be measured roughly by the platinum loop, or more carefully by a graduated capillary pipette. The disadvantages in the use of serum are entirely due to the slight difficulty in collecting and transporting it, and the delay in obtaining it when a blister is employed. If the serum is obtained from blood after clotting has occurred, a greater quantity of blood must be drawn than is necessary when the dried blood method is used. If it is obtained from a blister, a delay of eight to eighteen hours is required. The transportation of the serum in capillary tubes presents no difficulties if tubes of sufficiently thick and tough glass are employed and a proper case supplied.

THE TYPHOID CULTURE EMPLOYED. The majority of tests have been made with an eighteen, to twenty-hour broth-culture of a typhoid bacillus which was received from Pfeiffer during the past summer. Its virulence is such that one-fifth of a milligramme of a fresh agar-culture injected into the peritoneal cavity of a guinea-pig kills it in thirty-six to seventy-two hours.

Two other cultures of about one-half the virulence of the Pfeiffer culture have also frequently been used, and occasionally some one of a half-dozen other cultures whose virulence has not been tested. These latter include the culture used by Wyatt Johnston, which was kindly furnished to us. All these cultures gave fair reactions; some seemed to give more typical clumping than others, and the Pfeiffer culture particularly gave the most characteristic reactions. We have by chance used the method, adopted by Johnston, of planting our broth-cultures from old-stock cultures, but have not found it possible to avoid by this method moderate or pseudo-reactions in some cases not having typhoid fever. It seems to us that the main requirement is to have a broth-culture not over twenty-four hours old, in which the bacilli are isolated and actively motile, and the degree of whose motility is known.

THE DILUTION OF THE BLOOD OR SERUM TO BE EMPLOYED AND THE TIME REQUIRED FOR THE DEVELOPMENT OF THE REACTION. The serum-test, as shown by Widal, Grünbaum, and others, is quantitative and not qualitative. By this it is not meant to assert that the agglutinating and immobilizing substances produced in the blood of a typhoid patient are the same as those present at times in normal blood or those produced in the blood of persons sick with other diseases. It is intended, however, to maintain that the effect upon the bacilli as observed under the microscope is identical; the difference being that in typhoid fever, as a rule, the reaction occurs after the addition to the culture of far smaller quantities of serum than in other diseases, or when the same dilution is used it occurs far more quickly and completely with the typhoid serum. That the test is quantitative and not quali-

tative is well shown by the results of the examination of the seven following consecutive cases from Bellevue Hospital. Four of the cases were ultimately proved not to have been typhoid fever.

Case.	History, symptoms, and diagnosis at time of taking blood-specimens.	Corrected diagnosis on completion of illness.	Reaction of bacilli in broth-cultures to serum in different dilutions.		Reaction.
			Amount of serum.	Amount broth-culture	
1	Adult; sick four weeks, continuous high fever; pleurisy; "tuberculosis" with possibility of typhoid.	Tuberculosis.	1	1	Not appreciable.
2	Boy; sick two weeks; continued moderate fever, abating when test was made; prostration, constipation; no typhoid symptoms except fever and prostration; "atypical typhoid fever."	Uncertain.	1	1	Not appreciable.
3	Adult; symptoms of acute articular rheumatism only; "acute articular rheumatism."	Acute rheumatism.	1 1 1	1 4 9	Delayed moderate. Delayed very slight. Not appreciable.
4	Adult; just convalescent after sickness giving characteristic symptoms and physical signs of pneumonia; "pneumonia."	Pneumonia	1 1 1 1	1 4 9 19	Immediate marked. Delayed moderate. Delayed slight. Not appreciable.
5	Adult; continued high fever; enlarged spleen; typhoid bacilli obtained from spleen; "typhoid fever."	Typhoid fever.	1 1 1	1 4 9	Immediate. Delayed incomplete. Delayed very slight.
6	Adult; relapse after four wks of continuous fever with typhoid symptoms; "relapse after typhoid fever."	Typhoid fever.	1 1 1 1 1	1 10 50 100 200	Marked immediate. Marked immediate. Marked immediate. Delayed moderate. Delayed slight.
7	Adult; seven days continued high fever; typhoid symptoms; two days later an atypical rash; "typhoid fever."	Typhoid fever.	1 1 1 1 1	1 9 49 99 199	Marked immediate. Marked immediate. Marked immediate. Delayed but marked. Delayed moderate.

If other selected cases were added, a complete graduation in the intensity of the reaction with typhoid serum could be given. These seven suffice for the present purpose. It will be noted that two of the cases showed no reaction; neither of these had typhoid fever. A third gave a delayed moderate reaction with equal parts of the serum and culture; a fourth gave with this dilution an immediate marked reaction, and with one part to ten a delayed, slight reaction. No one of these four cases (which were not typhoid fever) gave an immediate marked reaction with a one in ten dilution, but one did give a delayed reaction. Of the typhoid cases one gave a reaction not more marked than that found in one of those not typhoid fever. The other two typhoids gave a characteristic reaction in a dilution twenty times greater than the others.

Our results have shown that in a certain proportion of cases not typhoid fever there occurs a delayed moderate reaction in a one in ten dilution of serum or blood; but very rarely, excepting in typhoid fever, does a complete reaction occur in this dilution within fifteen minutes. When dried blood is used the slight tendency of non-typhoid blood in one in ten dilution to produce agglutination is increased by the presence of the fibrinous clumps and perhaps by other substances derived from the disintegrated blood-cells.

The mode of procedure now employed is as follows: the test is first made with a one in ten dilution of serum or dried blood; if complete clumping and immobilization of the bacilli immediately occur, this is denominated "marked immediate typhoid reaction." The test is then repeated with greater dilutions.

If upon examination of the mixture in a hanging drop the bacilli show an immediate inhibition in their motility and a tendency to clump, and if upon later examination this clumping becomes marked within fifteen minutes, this is called a "moderate probable typhoid reaction." If within five minutes no change is noted in the motility of the bacilli, but slight clumping occurs later, this is noted for future reference and for its negative value; but such a reaction is never called a "typhoid reaction."

In order to determine whether the agglutinating substances are ever completely absent in typhoid fever, those cases in which no typhoid reaction has occurred in the usual one in ten dilution are retested with less dilutions of the serum. The complete absence of a reaction with equal parts of serum and broth-culture may be found to possess much greater value in excluding typhoid fever than a negative result obtained with the one in ten dilution.

THE PROPORTION OF CASES OF TYPHOID FEVER IN WHICH A DEFINITE TYPHOID REACTION OCCURS, AND THE TIME OF ITS APPEARANCE. In 108 cases in which, after the completion of the illness, a definite diagnosis of typhoid fever was finally made by the physician in charge, a marked immediate reaction occurred at the first test in 75. If the cases are arranged according to the day of the disease upon which the specimens were collected for examination, the following results are obtained:

Specimens collected.	Number of cases examined.	Reaction.		Percentage of positive results.
		Positive.	Negative.	
From 3d to 7th day	19	12	7	63 per cent.
During 2d week	39	23	16	59 "
" 3d week	24	19	5	79 "
" 4th week	9	8	1	88.8 "
" 2d month	17	13	4	76 "
	108	75	33	70 per cent.

The possibility that some of the cases giving no reaction and considered as typhoid fever by the physicians in attendance may not have had typhoid fever must be taken into consideration, yet many of these presented histories considered as fairly typical of typhoid fever. It is really not so important whether the reaction is present in 70, 80, or 90 per cent. of cases upon the first examination as that it is occasionally or frequently altogether absent.

In seventy-seven specimens from hospital cases in which the diagnosis was considered as almost certain, and in many of which several tests were made, there was a marked or definite typhoid reaction at some time in sixty-eight and only a slight reaction in nine. The reaction was never absolutely wanting. In hospital cases, therefore, in which repeated examinations were possible, a definite typhoid reaction was present in 88 per cent. of the cases at some time during the illness.

The majority of the cases that gave no reaction by the fourteenth day failed to give at any time a definite reaction; there were, however, two instances in which the reaction did not manifest itself until the middle of the fourth week, when the temperature had nearly reached the normal point.

The following case, in which numerous examinations were made, and which not only gave the symptoms of typhoid fever, but from the blood of whose spleen typhoid bacilli were obtained, is of great interest as proving that the blood of a typhoid patient may never, throughout the whole illness, have sufficient agglutinating substances in it to give a definite typhoid reaction in the usual dilution.

W. L., aged twenty years, Bellevue Hospital. No previous history of typhoid fever. Three weeks before admission had severe chill; during past two weeks has had continuous fever, and on admission presented the characteristic symptoms of typhoid fever. The temperature ranged between 103° and 104° for about ten days, and then fell to normal. Ten days later relapse occurred, which still continues at the time of writing, after the lapse of one week. Typhoid bacilli were obtained from the spleen by puncture on the twenty-fourth day. In this case throughout the first attack the reaction was never marked in a dilution above one part of serum to four of culture. On the second day of the relapse, however, the reaction became marked upon the addition of one part of serum to fifteen of culture.

If to the cases already tabulated are added those in which the duration of the disease at the time of the examination was not known, but in which the diagnosis of typhoid fever was established or excluded with considerable certainty, the following results are obtained:

	Reaction positive in one in ten dilution.	Reaction negative.
Number of cases finally believed to be typhoid	140	100
Number of cases finally believed not to be typhoid	57	0
Number of cases remaining in doubt, but regarded by the clinical observers as probably not typhoid	3	3
Total number of cases of suspected typhoid fever examined	200	

The period of time between the infection and the appearance of the reaction is well shown by the experimental inoculation of man and animals with small amounts of dead typhoid bacilli.

Following the experiment performed by Pfeiffer, in which he inoculated three healthy adults with small amounts of dead bacilli, we injected nine guinea-pigs with one-tenth of a loop of typhoid bacilli killed by exposure for one hour to heat at 55° C. The blood of these pigs was examined daily or on every alternate day by Dr. A. W. Williams, Assistant Bacteriologist to the Health Department, for the appearance of the typhoid reaction. As in Pfeiffer's experiments on man, the agglutinating substances did not appear in any experiments until the sixth day, and in all it appeared by the eighth day. The agglutinating substances gradually diminished, but were still present in appreciable quantity at the end of four weeks.

THE PERSISTENCE OF THE REACTION. The following table shows the results as to the persistence of the reaction in the cases examined :

Convalescent.	Cases.	Reaction.
3 to 4 months	8	Marked in 3, moderate in 2, very slight in 3.
1 year	1	Very slight, not definite typhoid.
3 years	1	None.
5 years	1	Very slight.
14 years	1	None.

The reaction disappeared on the thirty-fifth day in one case in which it had been marked on the thirtieth day.

THE REACTION WITH THE BLOOD-SERUM OF HEALTHY PERSONS AND OF THOSE SICK WITH DISEASES OTHER THAN TYPHOID FEVER. The reaction has never been observed in a one in ten dilution of the blood-serum of over one hundred healthy persons examined. Fifty-three persons have been examined who were regarded as having typhoid fever when the specimens of blood were collected, but in whom the subsequent history or the autopsy practically excluded this diagnosis. In no one of them did the blood give a marked immediate reaction in a one in ten dilution. Several gave a delayed partial reaction. These

cases, if divided according to the duration of illness when the examination was made, become grouped as follows:

Number examined during 1st week	.	.	.	.	.	13
" " " 2d "	.	.	.	.	.	27
" " " 3d "	.	.	.	.	.	6
" " " 4th "	.	.	.	.	.	4
" " " 2d month	.	.	.	.	.	3
						53

Besides these there were three cases which had a continuous, irregular temperature, but no other symptoms of typhoid fever. The blood of two of these gave an immediate definite reaction in dilution one in ten with the broth-cultures of the typhoid bacillus. The third gave an immediate moderate reaction.

These cases were treated at the Roosevelt and Bellevue Hospitals. In no one of the three was the nature of the illness determined. They were all colored adults. The histories of the cases are as follows:

CASE I.—Charlotte B., aged forty-five years, colored. Patient so ignorant as to make previous history untrustworthy. No history of previous typhoid obtained. For a few months she has had attacks of abdominal pain. Present illness began three days ago with nausea, vomiting, and pains in right iliac fossa. Physical examination on admission showed tenderness most marked in right iliac region; no tumor discovered. Spleen not noticeably enlarged. Temperature on admission 103.6° , and varied afterward between 103° and 104° for four days, then fell gradually during six days to 98.4° . Pulse varied from 90 to 105. Respirations 36 to 24. Three days after fall of temperature patient had a relapse, lasting a week. Temperature reached 104° F. on the third day. After partial subsidence of temperature an operation was performed for a possible appendicitis, but an inoperable tumor of the ascending colon was found. After the operation the temperature gradually subsided to normal and has remained so up to the present time, a period of two months, notwithstanding the continued growth of the tumor. Bowels were slightly costive throughout the illness. On the fifth day of her illness a solution of dried blood when mixed with the typhoid culture gave an immediate, very marked reaction in dilution one in ten.

During the first week of convalescence the reaction continued to manifest itself, but at the end of one month it had disappeared. In this case the previous history is valueless. Of typhoid symptoms there were none except the continued high temperature. No reason for the elevation of temperature was discovered. The tumor of the intestines hardly seems to have been the cause of the continued fever or of the substances giving the typhoid reaction, as they both subsided in spite of the continued growth of the tumor.

CASE II.—Thomas I., colored, Bellevue Hospital. Patient has not felt well for four weeks. Has lost some flesh and feels prostrated. No headache; feet and ankles swollen and painful for a week; appetite poor; bowels costive. On admission: temperature 101.4° , pulse 100, respiration

32. Physical examination negative, except that there is some swelling and oedema of the feet and ankles, with slight tenderness on pressure. Spleen not noticeably enlarged. History after admission to the hospital: temperature gradually subsided to nearly normal in four days and remained with morning temperature between 98° and 99° and evening between 99° and 100° for five days. It then rose to 104° and remained high for one week, after which it gradually subsided to nearly normal and remained so with the exception of two transitory rises.

The first rise in temperature was accompanied by increased general tenderness and heat in ankles and feet. Two days later there appeared areas of anaesthesia in skin of feet and toes and superficial gangrene of portions of toes and left foot, but the pain had disappeared from ankles and feet and patient felt better. He gradually improved and made a good recovery. Results of examination of serum: blood examined by Dr. J. W. Brannan seven days after admission showed no definite reaction. The blood, examined on the second day of relapse and at frequent intervals until patient was discharged, gave marked immediate reaction in a dilution one in ten, and a delayed incomplete reaction in a dilution one in twenty.

CASE III.—Adult, colored. As the reaction in a dilution one in ten, though fairly marked, was not complete, only a brief history will be given. Patient stated that he had been well until three days before admission, when, after wrestling, he had coughed up blood. On admission his temperature was 103°, pulse 90, respiration 26. Excepting for some signs of bronchitis, no physical signs of disease were found. Spleen apparently not enlarged.

An irregular fever, with temperature averaging about 103°, continued for two weeks, after which it gradually subsided. Except for the prostration and fever, there were no characteristic typhoid symptoms. On admission the diagnosis was acute phthisis or typhoid fever. The signs of disease in the chest disappeared, and patient was discharged cured.

In addition to the examinations made of the blood of persons supposed to have typhoid fever, in eighty-seven cases examinations have been made of the blood of persons who were sick with other diseases, such as tuberculosis, rheumatism, appendicitis, malignant tumors, diphtheria, scarlet fever, measles, leprosy, meningitis, leukæmia, etc. The serum from only one of these gave a marked immediate reaction in a one in ten dilution, and in this case the final diagnosis was doubtful.

The differential diagnosis of typhoid and typhus fevers is often of very great importance in a sanitary way, and in the absence of an epidemic of typhus fever is often difficult or impossible. It therefore becomes of great importance to determine whether the serum-test for the diagnosis of typhoid fever will serve to differentiate these diseases. Typhus fever is endemic in Liverpool, and with the hope of determining this question we requested Dr. E. W. Hope, Medical Officer for Health of Liverpool, to forward some samples of typhus blood. He very kindly sent three samples in capillary tubes; as we were afterward informed, all from cases in the same family. Two of these showed no

reaction, even when the serum and culture were mixed in equal parts; one showed a marked reaction. The possibility of a mistaken diagnosis of this case is strongly suggested, although we are informed that the case was one typical of typhus fever. As no other data on this question are at hand, it must be left unanswered for the present.

THE EFFECT OF THE SERUM FROM THE BLOOD OF THOSE SUFFERING FROM TYPHOID FEVER UPON OTHER VARIETIES OF BACILLI. Achard has found that certain bacilli, which, though closely allied to the typhoid bacillus, are still to be clearly distinguished from it, show the same clumping when mixed with the serum of immunized animals as the typhoid bacillus.

He also describes three cases from which these bacilli were obtained in which the serum of the patients caused marked clumping of the typhoid bacilli in dilution not greater than one in ten.

We have found that a number of varieties of motile bacilli other than the typhoid bacillus are clumped by the serum from persons suffering from typhoid fever, even when the serum is used in quite high dilutions.

These bacilli are also clumped, but to a much less extent, with serum from some persons suffering from other diseases. These experiments, which have been carried on chiefly by Dr. Hiss, Assistant Bacteriologist to the Health Department, are still incomplete.

A careful study of all the cases examined has been made to determine whether the agglutinating substances are more likely to be present in cases of typhoid fever which run a typical course than in those which present an irregular type of the disease.

An analysis of the 140 cases detailed above shows that although a reaction is perhaps more frequently obtained in the characteristic cases, yet it is often very strongly marked in atypical forms of the disease.

ANALYSIS OF RESULTS. The clumping and immobilization of the typhoid bacilli in broth-cultures occasionally caused by the blood-serum of persons ill with diseases other than typhoid fever are apparently identical with those caused by the serum from cases of typhoid fever; the difference being that in the latter the agglutinating substances are usually present in far greater amount. As, therefore, the serum-test for the diagnosis of typhoid fever is quantitative and not qualitative, it becomes necessary that a large number of examinations be made before it can be determined to how great an extent these agglutinating substances may be present in non-typhoid blood.

The exact diagnostic value of the serum-test and its precise limitations can only be determined when this is known. It is possible, however, to arrive at a probable estimate of its value from the results already obtained. From our investigations it would appear that the agglutinating substances are present in the blood-serum of over two-thirds of the cases

of typhoid fever by the end of the first week, in sufficient concentration to give a reaction with broth-cultures of the typhoid bacillus when these are mixed in the proportion of one to ten. A reaction with this dilution does not occur in more than 2 per cent. of non-typhoid cases. The reaction occurs in a dilution of one part of serum to forty parts of a broth-culture in over one-half of the cases of typhoid fever by the end of the first week; with this dilution the reaction rarely, if ever, occurs when serum from other cases is used.

It seems probable, therefore, that a positive diagnosis may be reached in 50 per cent. of the cases of typhoid fever, and a probable diagnosis in half the remaining cases, by estimating the amount of the agglutinating substances in the blood.

The absence of the reaction in a one to ten dilution in any case in a single examination does not exclude the diagnosis of typhoid fever, but the absence of the reaction in a series of examinations in a single case has decided value in probably excluding this disease.

The absence of the reaction will be useful in excluding typhoid fever in doubtful local epidemics where several cases can be examined, and in determining the nature of some fevers of a doubtful character; for although in no single case can typhoid fever be excluded, yet if the reaction occurred in no one of several cases this would clearly indicate that as a class these were not of the nature of typhoid fever. It is possible, also, that further examinations may show that where undiluted serum is used the complete absence of reaction has great value in positively excluding typhoid fever.

The direct microscopical examination, in the hanging drop, of a broth-culture of the typhoid bacillus and serum or blood mixed in proper proportions is the best method of applying the serum-test for general diagnostic purposes because of the small amount of serum or blood required. This method also seems as reliable as any.

Dried blood and blood-serum have given practically identical results; but there is a decided advantage in the use of the serum, because it may be used in a more concentrated mixture with the broth-culture than the dried blood in solution, and the proportions in the mixture may be more accurately determined. With dried blood in solution, the reaction, to be of value, should be fairly developed within a few (ten) minutes. With serum a longer time may be allowed for the development of the reaction, as pseudo-reactions are less likely to occur.

The duration of persistence in the blood, after convalescence, of the substances giving the reaction, varies greatly in different cases. In a few instances these substances rapidly diminish in amount; in most cases, however, they remain for some months, and exceptionally to a moderate extent for years. The substances rarely, if ever, persist more

than a year in sufficient quantity to interfere with the diagnostic value of the test in a subsequent illness.

This test will apparently be of value in many doubtful cases of typhoid fever in which the diagnosis remains uncertain after the fifth day of the illness, and in the differential diagnosis of those irregular forms of the disease which it is often difficult to distinguish clinically from typhus fever: malarial fever, miliary tuberculosis, malignant endocarditis, septicæmia, etc.

While the test is passing through the experimental stage it is impossible to make such positive statements as to the diagnostic value to be attached to the presence or absence of the reaction in any given case as may, perhaps, be permitted at a later time.

Meanwhile all observers in reporting cases should carefully note the method of examination, the dilution of serum or blood employed, and the exact grounds upon which the diagnosis is based. Those cases in which exact pathological or bacteriological examinations are obtained will be of especial value.

It is probable that there are obscure forms of disease due to the typhoid bacillus not now recognized as typhoid fever, and also other febrile affections now considered as typhoid fever which are really of another nature; therefore, unless great caution is observed in drawing conclusions, the reaction will be stated to have been present in cases, which were not typhoid fever when in reality these cases were typhoid fever; and, on the other hand, the reaction will appear to have been too frequently absent in cases of typhoid fever, because some of the cases were really of some other nature.

The method of serum-diagnosis in typhoid fever is simple and easy of performance in the laboratory by an expert bacteriologist, but it cannot be recommended for routine employment by the practising physician as a clinical test.

We desire, in conclusion, to express our obligations to many physicians for numerous data furnished, and also especially to Drs. Tillinghast and Clarke, of Roosevelt Hospital, and Drs. Don, Bigelow, and Goldberger, of Bellevue Hospital. Valuable assistance was rendered in these investigations by many members of the laboratory staff of the Health Department, and particularly by Drs. Lambert, Williams, Hiss, Guerard, and Wilson, assistant bacteriologists.

BIBLIOGRAPHY.

- Pfeiffer and Kolle: Zeitschrift f. Hyg., Bd. xxi, p. 203.*
Pfeiffer: Deutsch. med. Wochenschrift, 1896, Nos. 7 and 8.
Pfeiffer and Kolle: Ibid., No. 12.
Pfeiffer: Centralblatt f. Bakt., 1896, xix, p. 593.
Max Grüber: Wien. kün. Wochenschrift, 1896, Nos. 11, 12, and 14.
Grüber: Münch. med. Wochenschrift, 1896, No. 9.

Herbert E. Durham: Proceedings of the Royal Society, January 23, 1896, vol. lix., No. 355. Abstract in Brit. Med. Journ. *Epilemie*, March 14, 1896.

Durham: Lancet, December 19, 1896.

Albert S. Grünbaum: Lancet, September 19, 1896, and December 19, 1896.

Sheridan Delépine: Medical Chronicle, October, 1896, and Lancet, December 5 and 12, 1896.

F. Vidal: La Presse Médicale, July 29, 1896; La Semaine Méd., July 31; and Journ. de Méd., 1896, p. 533.

Vidal and Sicard: Le Bulletin Méd., September 19, 1896.

Vidal: Lancet, November 14, 1896.

Achard and Bensaude: Bulletin Méd., 1896, p. 333; La Semaine Méd., p. 295.

Chantemesse, Achard, Vidal, Hayem, Courmont, etc.: La Semaine Méd., 1896, p. 363.

Dieulafoy: Bulletin Méd., 1896, p. 655.

Vedel: Ibid., p. 763.

Haushalter: Ibid., p. 769.

Theolen and Mills: La Clinique (Brussels), August 6 and September 3, 1896.

Bordet: Annales de l'Institut Pasteur, April, 1895.

Greene: New York Medical Record, November 14, 1896.

Johnston: New York Med. Journ., October 31, 1896; Brit. Med. Journ., December 5, 1896.

THE EFFECT OF ANÆSTHESIA UPON TEMPERATURE AND BLOOD-PRESSURE.¹

BY DUDLEY P. ALLEN, M.D.,

PROFESSOR OF SURGERY IN THE MEDICAL DEPARTMENT OF WESTERN RESERVE UNIVERSITY, CLEVELAND, OHIO; VISITING SURGEON TO LAKESIDE AND CHARITY HOSPITALS.

THIS paper is based upon observations made upon thirty-five dogs and eighty patients. To these have been added the conclusions which seem to be warranted by the observations.

Before beginning to detail the experiments it may be desirable to state what led to their being undertaken. An extended observation of most of the noted surgeons, both of Europe and America, led me to question the special causes of their success or failure. Without considering the subjects of sepsis and antisepsis, there were three factors which particularly attracted my attention. Some surgeons were very slow in their operations, and seemed to disregard the loss of heat during prolonged operations upon patients with inadequate covering. I have seen one of the most noted surgeons of Germany perform a laparotomy lasting nearly two hours, the patient covered only with a couple of sheets, in a room with the window open, in which it was uncomfortably cool for me while watching the operation, clad in a winter suit. Another surgeon of equal reputation performed all his laparotomies with the patient lying upon a glass slab covered only with a sheet. On the other hand, I have seen another surgeon, equally noted, much less careful in his asepsis, but still very successful. He exercised, however, the greatest care to prevent loss of time and loss of blood, and to keep the patients well protected and warm.

¹ Read before the American Surgical Association, 1896.